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Reproductive Toxins and Alligator Abnormalities at Lake Apopka, Florida

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The alligator population at Lake Apopka in central Florida declined dramatically between 1980 and 1987. Endocrine-disrupting chemicals and specifically DDT metabolites have been implicated in the alligators' reproductive failure. The DDT metabolite hypothesis is based largely on the observation of elevated concentrations of *p,p*-DDE and *p,p*-DDD in alligator eggs obtained from Lake Apopka in 1984 and 1985. In the following commentary, we draw attention to two nematocides that are established reproductive toxins in humans, dibromochloropropane (DBCP) and ethylene dibromide (EDB), which could also have played a role in the reproductive failure observed in alligators from Lake Apopka in the early 1980s. *Key words*: alligator, DBCP, DDT, EDB, environmental estrogen, nematocides, pesticides, reptile.
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Developmental abnormalities of the gonad and abnormal sex hormone concentrations have been documented in juvenile alligators from Lake Apopka (*1*). Recently, it has been shown that many of the environmental chemicals found in alligator plasma or eggs bind the alligator estrogen and/or progesterone receptors *in vitro* (*2*). Guillette et al. (*1*) suggested that the reproductive failure at Lake Apopka could have been related to general agricultural pollution and to a spill from a nearby pesticide manufacturing facility. From 1957 to 1981, the facility (Tower Chemical Co.) manufactured and stored both chlorinated and organophosphate insecticides as well as a copper–salt-based fungicide at a site 1.5 miles from Lake Apopka. Wastewater from the manufacturing process was discharged into an unlined pond, and chemicals were burned or buried on site. During a heavy rain in 1980, the percolation pond overflowed and acidic wastewater discharged into a marsh that drains into Lake Apopka. DDT and other chemicals contaminated the lake during this extensive spill. The area surrounding the chemical company's plant was declared an EPA Superfund site in 1983. DDT and other pesticides have also entered the lake as a result of extensive agricultural activity surrounding the lake, primarily activity in orange groves and vegetable muck farms. Because of the endocrine-disruptive potential of DDT's degradation products DDE and DDD, they have been the prime suspects in the reproductive abnormalities of the alligators (*1*).

During a review of sampling reports and inventory documents, we became aware of two other reproductive toxins, 1,2-dibromo-3-chloropropane (DBCP) which is known by the trade name Nemagon, and ethylene dibromide (EDB), that could also have contributed to a decline of the alligator population. DBCP, a well-established human reproductive toxin, was first reported to be associated with male sterility at a California pesticide-formulation plant in 1977 (*3*). The EPA banned DBCP from all U.S. crops in 1985, and it was subsequently replaced by EDB. Since 1948, EDB has been used extensively as a fumigant for grain, fruit, and vegetable infestations. EDB has also been shown to be a reproductive toxin and to damage sperm and decrease fertility in humans (*4*). Both of these chemicals were present in the Lake Apopka environment at elevated concentrations according to historic monitoring records described below.

Dibromochloropropane

In addition to being contaminated by DDT and its metabolites, the soil, sediment, and surface water at the Superfund site also contained high levels of DBCP following the spill. The method of waste disposal used by the company was to burn and bury waste in an area of approximately 58,200 ft^2, from which up to 70 drums were excavated during clean-up efforts. Analytical results from the 1980 EPA (*5*) and the 1981 Florida Department of Environmental Regulation (FDER) (*6*) sampling investigations, as well as analyses of material uncovered during the 1983 clean-up of the burn/burial site, confirm that pesticide waste was disposed of at that site. DBCP was found at concentrations as high as 1,340,000 ppb in surface soil samples at the burn/burial site (Table 1) (*5–7*). In 1983, portions of the burn/burial area were removed by the EPA. DBCP was also found in sediment samples collected from the wastewater pond by the EPA and the FDER in 1980, 1981, and 1983 in the range of 162,000–165,000 ppb (Table 1) (*5–7*). Subsequently, the pond was drained and sediment, to an average depth of 2 ft, was removed from an area estimated to be 30,100 ft^2. However, the remaining lower sediment layers of the former wastewater pond remain a potential source of contaminants, subject to groundwater migration. Surface water samples taken in 1983 from the lagoon and a stream off-site contained only low levels of DBCP contamination (<0.041 ppb) (*7*). This finding is consistent with modeling data using the Henry's law constant, which indicates a half-life of 13.5 hr for evaporation from a model river 1-m deep (*8*). The physicochemical properties of DBCP are very different from those of DDT in that DBCP is very volatile and does not bioaccumulate (*9,10*). A volatilization half-life of 8 days can be calculated from a model pond by using a three-compartment EXAMS model (*11*). The high environmental mobility of DBCP in soil and water (*12*) may have contributed to the fact that it was not detected in subsequent samples and that the EPA decided not to include it on the short list for long-term monitoring.

The route of exposure for the alligators living in this environment might have occurred through both oral and dermal exposure. Animal studies in rats show that DBCP is rapidly absorbed from the gastrointestinal tract after oral administration by gavage when water is used as a vehicle (*13*). Dermal absorption is not as well documented in animal studies, but it is supported by the observation that death occurred following a 24-hr dermal exposure (*14*). Occupational exposures to DBCP probably occurred through both inhalation and dermal exposure; thus, the contribution of dermal absorption to reproductive health outcomes cannot easily be determined.

The testicular abnormalities in alligators from Lake Apopka are similar to those in pesticide workers exposed to DBCP in that the seminiferous tubules are the affected target tissues. However, whereas alligator testes

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show poorly organized seminiferous tubules with bar-shaped cellular structures (*1*), the seminiferous tubules from humans exposed to DBCP are devoid of spermatogenic cells altogether (*15*). Moreover, the mechanism by which these effects occur may be different. Guillette et al. (*16*) have proposed that the developmental abnormalities of the gonad reported in alligators is due to organizational modifications; that is, the development of the testis has been altered such that the molecular and/or cellular functions of this organ are modified. Thus, the changes observed are permanent and not reversible. In contrast, the observed testicular abnormalities in pesticide workers are activational modifications affecting the same organ and apparently similar cell types—the spermatogonia. Among pesticide workers diagnosed with oligospermia, normal testicular functioning was restored once the workers were removed from the source of DBCP because their testes were organizationally normal. It is possible that the depressed clutch viability and reduced neonatal survival among these alligators (*1*) parallel the increase in spontaneous abortions observed among humans following exposure to DBCP (*17*); that is, exposure of alligators to DBCP following the spill may have modified male reproductive functioning dramatically by an activational mechanism, resulting in poor recruitment and thus a population decline. A reduction in male alligator reproductive ability in concert with altered female reproductive functioning and elevated persistent, biomagnified contaminants in eggs (such as DDT metabolites) would lead to the dramatic population decline and egg/juvenile mortality reported (*16,18*).

Extensive monitoring initiated in 1983 by the Florida Department of Environmental Protection (FDEP) and Florida Department of Health and Rehabilitative Services (FHRS) failed to reveal DBCP contamination in ground water in wells surrounding the lake (C. Cosper, personal communication; A. Reich, personal communication). However, given the levels found in the remaining pond, it is almost certain that DBCP entered Lake Apopka during the 1980 spill.

Ethylene Dibromide

Sampling of private wells initiated in 1984 in the vicinity of Lake Apopka by the FHRS found another reproductive toxin, EDB, at concentrations exceeding the Florida maximum contaminant limit (MCL) of 0.02 ppb. The majority of sampling by the FHRS in this area was conducted in the southwest corner of the lake where the Superfund site is located. More than 80 wells were found to have EDB levels above the MCL; 29 of these wells were contaminated with levels greater than 1.0 ppb (Fig. 1). EDB was a commonly used agricultural nematocide in Florida; therefore, the contamination is likely to have originated from agricultural use rather than the spill. Because the EDB-contaminated wells are located upstream from the Lake Apopka aquifer, which regionally flows toward the northeast, cross-contamination of the lake could have occurred. In field experiments, EDB has been detected in soil 19 years after its last known application (*19*). In aquifers, where volatization does not occur, the half-life for EDB undergoing uncatalyzed hydrolysis is 6 years (*20*). Like DBCP, EDB is very volatile and does not bioaccumulate (*21,22*); EDB can be absorbed via the dermal or oral routes (*23*). In humans, EDB is known to be associated with decreased sperm count, an altered percentage of viable and motile sperm, and an increase in certain types of morphological abnormalities of sperm (*4,24*). If this compound was present in the lake in the early 1980s, which is very likely, and given its use in agricultural activities and presence in samples from wells surrounding the lake, it could also have contributed to the reproductive problems that led to a decline in the alligator population. However, to our knowledge, no monitoring of EDB at the Superfund site was conducted on water, muck, or biological samples from the lake during the early 1980s. Therefore, historic inferences cannot be drawn from current monitoring efforts.

Table 1. Concentrations of 1,2-dibromo-3-chloropropane (DBCP) detected in samples taken by the EPA and the Florida Department of Environmental Regulation at the Superfund site near Lake Apopka

Site	Concentration of DBCP (ppb)	Reference
Surface soil samples from burn/burial area (0–1 ft)	1,337,792	(*5*)
Surface soil samples from burn/burial area (1.0 ft)	10–1,500	(*7*)
Surface soil samples from burn/burial area (2 in)	10.0–750	(*7*)
Sediment samples from waste water pond (1 ft)	162,000–165,000	(*5–7*)
Sediment samples from waste water pond (2 ft)	200,000–300,000	(*7*)
Surface water samples from the unnamed stream	<0.041	(*7*)

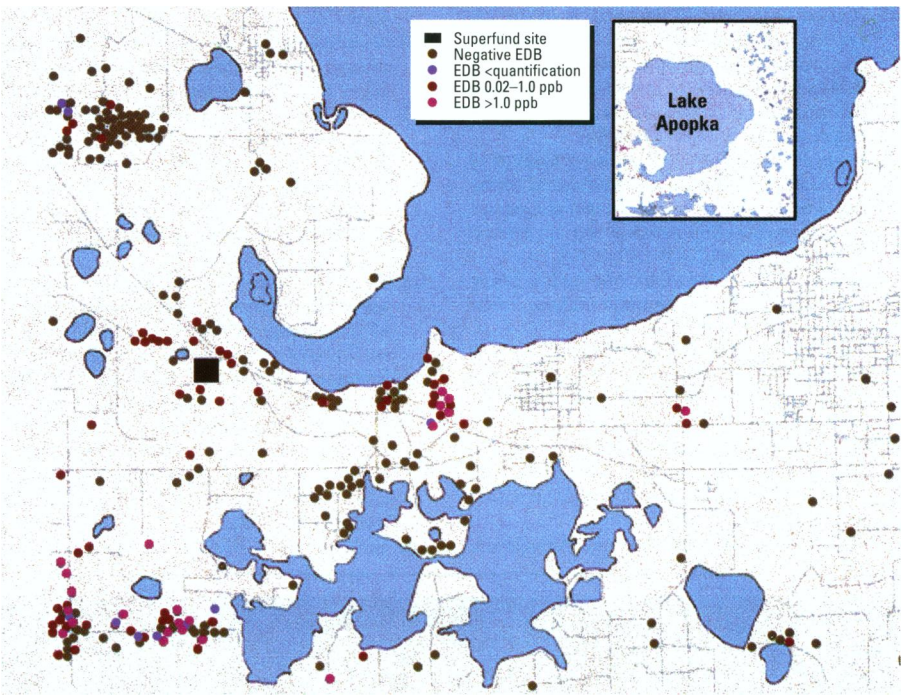

Figure 1. Private well sampling and ethylene dibromide (EDB) testing in the vicinity of Lake Apopka (insert), Florida. The Florida maximum contaminant limit is 0.02 ppb (µg/l), with remediation of wells occurring at levels greater than 0.025 ppb. Data were obtained from the Florida Department of Health and Rehabilitative Services (unpublished results).

Conclusion

Although DDT, its metabolites, and other persistent bioaccumulated pesticides continue to be prime suspects in the egg and embryonic abnormalities reported, the alligator population decline that occurred in the early 1980s at Lake Apopka could have involved two other potent reproductive toxins as well. DBCP and EDB, two documented reproductive toxins in humans, were observed in elevated concentrations at the

Superfund site (DBCP) and in well water (EDB) and could have affected alligators at Lake Apopka.

The findings discussed above indicate a complex exposure scenario in which the etiology of the reproductive failure cannot be reconstructed with certainty due to the historic nature of the event.

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